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CD133免疫磁珠分选脐血内皮祖细胞的培养及鉴定

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摘要：

目的: 建立具备高效增殖、血管生成与迁移能力的脐血内皮祖细胞（endothelial progenitor cell，EPC）分离培养鉴定方法。方法: 应用免疫磁珠分选纯化脐血单个核细胞中的CD133　+细胞，体外EGM 2 MV培养液培养扩增，通过形态学、细胞表面标志及细胞功能鉴定EPCs，并与脐静脉内皮细胞（human umbilical vein endothelial cell，HUVEC）作比较。结果: EPCs分离培养7 d左右开始出现小集落，21 d左右集落扩大、相互融合，并呈现出典型铺路石样改变；培养14 d左右，约90%的细胞免疫荧光DiI ac LDL和FITC UEA 1双阳性，阳性率达90%。CD133 和CD34 阳性率从86.04%降至2.96%、90.88%降至2　99%，而CD31阳性率从1.12%升至99.88%。在增殖、血管生成与迁移能力上比较，EPCs明显优于HUVECs（　P　<0.05）。结论: 通过CD133免疫磁珠分选脐血单个核细胞，可培养出具有高效增殖、血管生成与迁移能力的EPCs。

关键词：内皮祖细胞 CD133 免疫磁珠分选 增殖 血管生成 迁移

Culture and identification of endothelial progenitor cells from cord blood with CD133 immunomagnetic sorting　Download Fulltext

Fund Project:Supported by the Medicine Science and Technology Development Foundation of Jiangsu Health Department (No.H200510); the Medical Major Talent Program of Jiangsu Province（No.RC2007061）

Abstract:

Abstract Objective: To establish a method for isolating and culturing endothelial progenitor cells（EPCs), which have high potential of proliferation, migration and angiogenesis, from cord blood. Methods: CD133　+cells were selected from fresh cord blood mononuclear cells by magnetic activated cell sorting system (MACS), and were cultured in EGM　2MV　medium. EPCs were identified by examining the morphology, cell markers and functions. And the EPCs were compared with human umbilical vein endothelial cells (HUVECs). Results: On the 7　th　day, the adherent cells exhibited the small colonies; and on the 21　th　day,the colonies were expanded, confluenced and displayed a typical “cobblestone” morphology. On the 14　th　day, 90% attached cells took up DiI ac LDL, and bound FITC UEA 1 (double positive fluorescence). The cell markers of CD133 and CD34 decreased from 86.04% to 2.96% and 90.88% to 2.99%, respectively, while CD31 increased from 1.12% to 99.88%. Compared with HUVECS, EPCs had more potent potential of proliferation, migration and angiogenesis(　P　<0.05). Conclusion: CD133　+ MACS can be used to isolate EPCs, with high capacity of proliferation, angiogenesis and migration, from cord blood mononuclear cells.

Keywords:endothelial progenitor cell CD133 immunomagnetic sorting proliferation angiogenesis migration

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